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Translate This … during Dietary Restriction

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Several studies indicate that reduced TOR signaling underlies life span extension by dietary restriction. Recently, Zid et al. (Zid et al., 2009) linked the benefits of dietary restriction in flies to increased levels of the downstream TOR target 4E-BP1 and corresponding changes in the relative translation rates of classes of mRNAs.

Dietary restriction (DR), reduced nutrient intake without malnutrition, extends life span in model organisms from yeast to primates, yet the mechanism(s) that underlie these effects remain a matter of debate. Several reports implicate reduced activity of the nutrient-responsive kinase TOR (target of rapamycin) as a major beneficial effect of DR (see Stanfel et al., 2009 for review and references therein). Thus, a central question is which downstream effectors of TOR signaling are linked to aging. The best-characterized TOR substrates, ribosomal protein S6 kinases (S6Ks) and 4E-BPs (eIF4E-binding proteins), both control translation initiation and are implicated in aging. S6Ks phosphorylate several translation-associated substrates, including ribosomal proteins and factors involved in initiation and elongation. Reduced S6K activity confers life span extension in yeast (*S. cerevisiae*), worms (*C. elegans*), and flies (*D. melanogaster*). When active, 4E-BPs bind to eIF4E, obstructing its interaction with eIF4G and inhibiting translation. Reduced eIF4E activity leads to life span extension in worms. TOR phosphorylation activates S6K and inhibits 4E-BP, with both events stimulating translation initiation; therefore, through reduced TOR activity, DR dampens translation initiation. Although other TOR substrates exist, S6Ks and 4EBPs are prime candidates for factors downstream of TOR that modulate aging.

In a recent *D. melanogaster* study, Zid et al. explore the link between DR and 4E-BP (Zid et al., 2009). Their results confirm that DR, diminished levels of yeast extract, results in reduced protein translation. Interestingly, they also detected increased protein levels of d4E-BP, the sole fly 4E-BP ortholog. In longevity studies, *d4E-BP* null flies are short-lived on rich food and refractory to DR-mediated life span extension. Furthermore, overexpression of *d4E-BP*-activated alleles result in life span extension on rich food sources but does not further extend dietary-restricted flies. Therefore, a primary effect of DR may be enhanced d4E-BP activity, raising the critical question of how DR evokes this response. Do enhanced levels of d4E-BP result from diminished TOR activity, or is there another route linking reduced nutrient load to d4E-BP?

While decreased translation initiation may underlie the longevity benefits of reduced TOR signaling, turning down TOR also enhances autophagy and other stress-response pathways possibly impacting aging. To measure translation changes, Zid et al. (Zid et al., 2009) employed microarray-based translation state array analysis (TSAA) (Arava et al., 2003; MacKay et al., 2004) to determine global changes in ribosome occupancy of mRNA species (denoting changes in translation efficiency) during DR as compared to flies on rich food. In the Zid et al. study (Zid et al., 2009), changes in translation efficiency for each mRNA were estimated from the proportion associated with <5 ribosomes versus 5 or more.

Reduced expression of ribosomal protein genes extends life span in yeast and worms, leading to speculation that a general reduction in protein synthesis may be beneficial, perhaps due to a corresponding reduction in protein damage with age. Findings from Zid et al., however, indicate concerted changes more indicative of a coordinated alteration in the corresponding proteome (Zid et al., 2009). Yeast replicative life span extension by DR also involves specific, albeit different, changes in translation (Steffen et al., 2008). In the DR fly study, an inverse relationship between the relative translation level and 5′UTR complexity was discovered; messages with simple 5′UTRs had relatively more ribosome occupancy than those with complex 5′UTRs. Among the classes of genes with increased ribosome loading were electron transport components, particularly those associated with complexes I and IV, leading to the hypothesis that DR induces mitochondrial activity. This was confirmed by measuring complex I and cytochrome C oxidase activities and linked to the aforementioned increase in d4E-BP protein levels, since the increase in mitochondrial activity was abrogated in *d4E-BP* null flies undergoing DR. An intriguing side note from the TSAA results is that many mRNAs with the greatest shift toward higher ribosome occupancy have relatively small coding sequences. Occupancy by five or more ribosomes for the smallest (coding sequence of 306 nt) would require nearly maximal ribosome packing. An earlier high-resolution TSAA study in yeast also found a high prevalence of small mRNAs, many encoding mitochondrial proteins, in fractions associated with ≥20 ribosomes (MacKay et al., 2004). Based on both studies, it may be premature to rule out an alternative explanation that a subcellular structure retains polysome association even under high detergent extraction.

The primary question for aging was whether enhanced mitochondrial activity was required for life span extension by DR. To test this, DR was performed on RNAi-treated flies with reduced complex I or complex IV activity (Zid et al., 2009). In both cases, reduced activity attenuated but did not completely block life span extension. Multiple possible interpretations exist for this partial impedance.

One is that the RNAi approach only modestly reduced complex I or complex IV activity, allowing the flies a partial response to DR. Alternatively, *d4E-BP*-dependent but oxidative phosphorylation-independent pathways may account in part for life span extension by DR. In any case, these findings suggest that a major benefit of DR derives from enhanced mitochondrial activity. In another recent paper using a Parkinson's disease fly model, Tain et al. (Tain et al., 2009) showed that increased activity of d4E-BP via rapamycin administration or direct overexpression suppresses pathological phenotypes, including muscle degeneration, reduced locomotor ability, and mitochondrial dysfunction. Overexpression of *d4E-BP* also protects against an age-related decline in fly cardiac function (Wessells et al., 2009).

Despite intense investigation, the relationship between mitochondrial function and aging remains enigmatic. Mitochondrial function declines with age in several organisms, making it feasible that DR extends life span by offsetting this decline through induced mitochondrial biogenesis (see Lopez-Lluch et al., 2008 for review). This may be conserved, since enhanced mitochondrial biogenesis is reported in mammalian DR studies. In contrast, in *C. elegans*, adult-specific RNAi to *reduce* expression of oxidative phosphorylation components results in life span extension (see Wolff and Dillin, 2006 for review). In yeast chronological aging, respiratory function is required for life span extension by reduced TOR signaling (see Stanfel et al., 2009 for review and references therein); however, this is not the case for replicative life span with reduced TOR signaling or DR. Similarly, efforts to extend life span by reducing reactive oxygen species have met with mixed success. One intriguing possibility is that enhanced mitochondrial function(s) independent of oxidative phosphorylation slow aging. Further experiments will be needed to resolve these questions.

Recent findings have emphasized the TOR pathway as a conserved modulator of aging. A quantitative study of conserved longevity determinants between worms and yeast identified several components linked to TOR signaling and translation (Smith et al., 2008). Moreover, rapamycin, a specific inhibitor of TOR signaling, was recently shown to extend life span in mice, even when the mice were not exposed to the drug until 20 months of age (Harrison et al., 2009). These findings raise the possibility that interventions in the TOR pathway may prolong human health span and make critical a mechanistic understanding of the pathways downstream of TOR linked to aging.

REFERENCES

Arava, Y., Wang, Y., Storey, J.D., Liu, C.L., Brown, P.O., and Herschlag, D. (2003). Proc. Natl. Acad. Sci. USA *100*, 3889–3894.

Harrison, D.E., Strong, R., Sharp, Z.D., Nelson, J.F., Astle, C.M., Flurkey, K., Nadon, N.L., Wilkinson, J.E., Frenkel, K., Carter, C.S., et al. (2009). Nature *460*, 392–395.

Lopez-Lluch, G., Irusta, P.M., Navas, P., and de Cabo, R. (2008). Exp. Gerontol. *43*, 813–819.

MacKay, V.L., Li, X., Flory, M.R., Turcott, E., Law, G.L., Serikawa, K.A., Xu, X.L., Lee, H., Goodlett, D.R., Aebersold, R., et al. (2004). Mol. Cell. Proteomics *3*, 478–489.

Smith, E.D., Tsuchiya, M., Fox, L.A., Dang, N., Hu, D., Kerr, E.O., Johnston, E.D., Tchao, B.N., Pak, D.N., Welton, K.L., et al. (2008). Genome Res. *18*, 564–570.

Stanfel, M.N., Shamieh, L.S., Kaeberlein, M., and Kennedy, B.K. (2009). Biochim. Biophys. Acta. *1790*, 1067–1074.

Steffen, K.K., MacKay, V.L., Kerr, E.O., Tsuchiya, M., Hu, D., Fox, L.A., Dang, N., Johnston, E.D., Oakes, J.A., Tchao, B.N., et al. (2008). Cell *133*, 292–302.

Tain, L.S., Mortiboys, H., Tao, R.N., Ziviani, E., Bandmann, O., and Whitworth, A.J. (2009). Nat. Neurosci. *12*, 1129–1135.

Wessells, R., Fitzgerald, E., Piazza, N., Ocorr, K., Morley, S., Davies, C., Lim, H.Y., Elmén, L., Hayes, M., Oldham, S., and Bodmer, R. (2009). Aging Cell, in press.

Wolff, S., and Dillin, A. (2006). Exp. Gerontol. *41*, 894–903.

Zid, B.M., Rogers, A., Katewa, S.D., Vargas, M.A., Kolipinski, M., Lu, T.A., Benzer, S., and Kapahi, P. (2009). Cell, in press.